

Supporting Information

LC-MS/MS-Based Method for the Multiplex Detection of 24 Fentanyl Analogues and Metabolites in Whole Blood at Sub ng mL⁻¹ Concentrations

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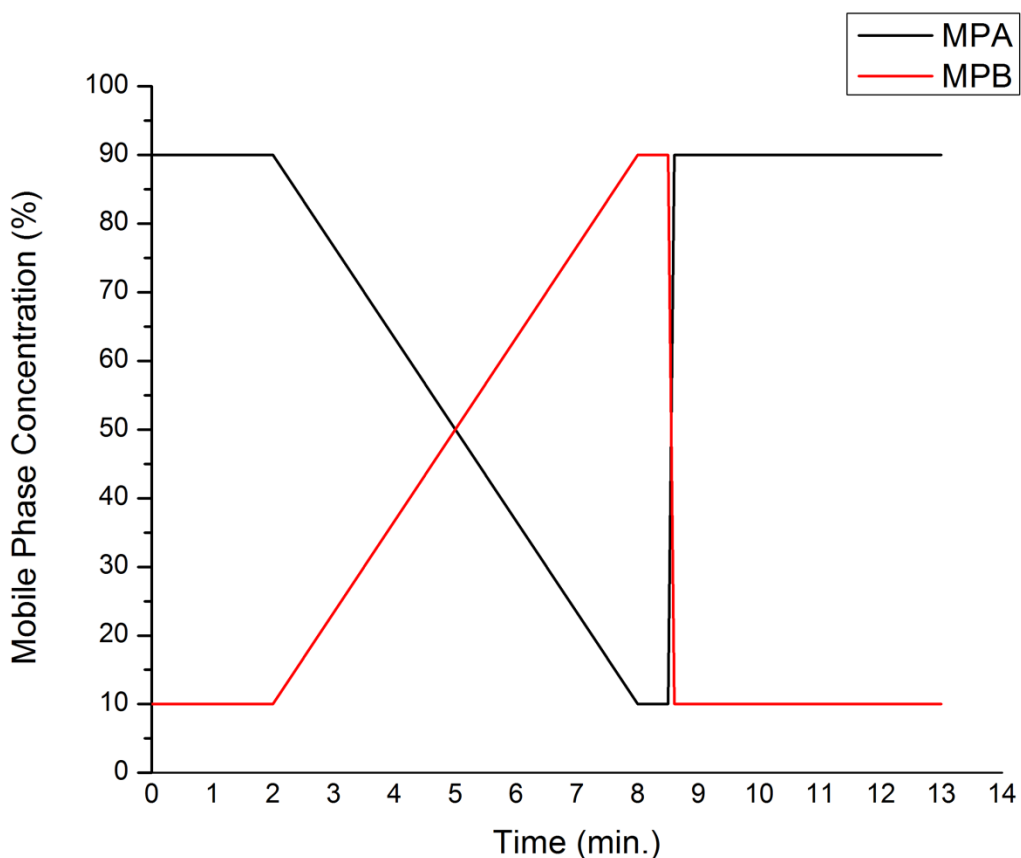


Figure S1. MPA and MPB gradient concentration change over a 13.5 minute per sample run period.

Table S1. Drugs commonly detected in toxicological screening ($N = 70$) and tested in this study for exogenous interferences.

Injection Mix	Drugs included in the interference study test mix	Concentration (ng mL ⁻¹)
Benzodiazepines	7-Aminoclonazepam, 7-Aminoflunitrazepam, Alprazolam, Chlordiazepoxide, Clonazepam, Diazepam, Flunitrazepam, Flurazepam, Lorazepam, Midazolam, Nordiazepam, Oxazepam, Phenazepam, Temazepam, Triazolam	2,500
Opiates	Hydrocodone, Hydromorphone*, Oxycodone, Morphine, Oxymorphone*, 6-Acetylmorphine*, Codeine	2,500* 10,000
Stimulants	Cocaine, Cocaethylene, Ecgonine methyl ester, Benzoyllecgonine, Amphetamine, Phenylpropanolamine, Phentermine, Methamphetamine, Pseudoephedrine, Methylenedioxyamphetamine, Benzyl piperazine, Methylenedioxymethamphetamine, Methylenedioxyethylamphetamine, Methylphenidate	1,250
THC	Tetrahydrocannabinolic acid*, Tetrahydrocannabinol	1,250* 500
Bases	Chlorpheniramine, Citalopram, Dextromethorphan, Doxylamine, Lidocaine, Methadone, Mirtazapine, Tramadol, Trazodone, Verapamil, Zolpidem	1,250
AD	Venlafaxine, Desvenlafaxine, Quetiapine, Paroxetine, Desipramine, Norfluoxetine, Imipramine, Fluoxetine, Nortriptyline, Cyclobenzaprine, Amitriptyline, Sertraline, Duloxetine	1,250
AE	Gabapentin, Lamotrigine, Levetiracetam, Pregabalin, Topiramate, Zonisamide	5,000
GHB	Gamma hydroxybutyric acid	50,000
Rx	Naloxone	10,000

Table S2. Recovery percentages for $N=22$ illicitly manufactured fentanyl (IMF) analogues and metabolites at low (0.35 ng mL^{-1}), medium (2.5 ng mL^{-1}), and high control concentrations (25.0 ng mL^{-1}). Set #1 and #2 refer to post extraction spiked and regular extraction samples, respectively.

Norfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	17631.48	140730.77	1000604.93		13814.81	108589.51	894437.11
2	18047.00	137202.97	829527.44		14056.68	89730.02	960464.26
3	18557.65	130331.00	1175213.13		16278.87	101081.64	872363.61
Average	18078.71	136088.25	1001781.83		14716.78	99800.39	909088.33
Recovery (%)	81.40	73.34	90.75				
Furanyl norfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	4590.15	31704.02	230975.70		2561.18	25373.77	203161.69
2	5487.52	28719.55	212752.81		2868.62	15808.62	254305.58
3	5446.13	27056.84	304847.16		3643.37	17184.88	197428.08
Average	5174.60	29160.14	249525.22		3024.39	19455.76	218298.45
Recovery (%)	58.45	66.72	87.49				
Remifentanil acid							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	2089.06	16561.54	167171.23		888.60	5835.51	50776.07
2	2334.54	16266.17	157158.90		800.58	4657.90	52331.12
3	2339.62	15863.99	155740.28		885.15	5417.15	49659.41
Average	2254.40	16230.56	160023.47		858.11	5303.52	50922.20
Recovery (%)	38.06	32.68	31.82				
Butyryl norfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	19330.65	177026.12	1153778.73		18740.57	135942.06	1035652.82
2	21813.09	175292.19	934465.45		17835.67	118133.80	1076581.04
3	20551.76	168768.33	1307357.39		21253.04	129095.32	992372.29
Average	20565.17	173695.55	1131867.19		19276.43	127723.73	1034868.72
Recovery (%)	93.73	73.53	91.43				
Remifentanil							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	3398.58	25879.27	247196.84		2769.84	19518.17	206515.06

2	3631.82	26098.03	251472.03		2732.97	16166.15	214967.39
3	3855.28	23094.96	262547.87		2863.09	20413.23	191769.16
Average	3628.56	25024.09	253738.91		2788.64	18699.18	204417.20
Recovery (%)	76.85	74.72	80.56				
Acetyl fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	39163.01	307276.32	2737509.47		32202.32	242612.47	2319607.20
2	39599.84	295551.80	2686140.35		31485.04	208628.84	2443187.71
3	41336.33	279207.70	2690796.28		35728.32	246516.70	2248135.27
Average	40033.06	294011.94	2704815.37		33138.56	232586.00	2336976.72
Recovery (%)	82.78	79.11	86.40				
Alfentanil							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	16613.52	111052.10	1069888.51		9410.82	83578.37	829356.47
2	16148.05	106762.53	1091804.94		10494.02	68625.62	847084.67
3	16345.64	96063.01	1030880.31		11468.14	85460.45	800546.70
Average	16369.07	104625.88	1064191.25		10457.66	79221.48	825662.61
Recovery (%)	63.89	75.72	77.59				
AH-7921							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	5425.16	41519.09	171578.91		4987.49	30028.21	268991.42
2	5448.88	38275.23	187181.59		4626.44	29397.38	299962.16
3	5587.03	35003.78	317460.45		4828.00	33563.29	248723.39
Average	5487.02	38266.03	225406.98		4813.98	30996.29	272558.99
Recovery (%)	87.73	81.00	120.92				
U-47700							
	Set #1				Set 2		
Trial no.	Low	Med	High		Low	Med	High
1	8789.37	69172.60	663875.37		7419.27	55748.60	525822.70
2	7814.93	65002.70	675335.12		7911.76	51056.55	554450.77
3	9579.90	60507.01	611724.71		8857.27	53830.68	501931.10
Average	8728.06	64894.10	650311.73		8062.76	53545.28	527401.53
Recovery (%)	92.38	82.51	81.10				
Acetyl fentanyl 4-methylphenethyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High

1	43670.09	362857.36	3004272.27		39273.94	294742.30	2564883.21
2	49639.28	358218.25	2896594.04		40145.21	270667.02	2671448.69
3	52373.23	325855.55	2865642.66		46585.53	276548.83	2533831.52
Average	48560.87	348977.05	2922169.66		42001.56	280652.71	2590054.47
Recovery (%)	86.49	80.42	88.63				
Acrylfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	46580.17	341583.14	2646397.46		37202.88	281712.87	2428913.55
2	46952.36	337096.36	2475688.79		37972.97	242770.34	2537661.55
3	48849.41	310714.29	2705321.61		43752.84	287579.53	2351749.58
Average	47460.65	329797.93	2609135.95		39642.89	270687.58	2439441.56
Recovery (%)	83.53	82.08	93.50				
Fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	36202.96	270883.19	2119693.09		29709.08	227320.04	2041829.06
2	35742.90	270642.79	2000594.92		29167.22	198024.25	2134377.46
3	36708.57	245358.38	2271119.18		35764.60	218804.82	1996871.66
Average	36218.14	262294.79	2130469.06		31546.97	214716.37	2057692.73
Recovery (%)	87.10	81.86	96.58				
<i>para</i>-Methoxyfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	27904.20	216676.89	1614255.82		22788.33	171129.57	1444155.49
2	28721.90	201953.30	1627239.48		22972.15	154837.11	1453530.64
3	27416.76	187643.49	1650187.28		28583.31	181306.70	1446219.28
Average	28014.29	202091.22	1630560.86		24781.26	169091.13	1447968.47
Recovery (%)	88.46	83.67	88.80				
4-ANPP							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	166174.74	1261945.01	8907584.39		190178.80	1229445.02	8369358.55
2	164486.85	1336291.01	7977401.72		180041.98	1076604.71	8607365.57
3	153677.98	1223611.13	9025869.40		202286.59	1208246.04	7867551.53
Average	161446.52	1273949.05	8636951.83		190835.79	1171431.92	8281425.22
Recovery (%)	118.20	91.95	95.88				
Furanyl fentanyl							
	Set #1				Set #2		

Trial no.	Low	Med	High		Low	Med	High
1	52810.26	387507.42	2941574.99		43119.60	311988.90	2601610.85
2	55872.20	383238.32	2976957.89		42896.47	273618.10	2713430.81
3	54758.43	355667.52	3015865.39		49315.64	319827.23	2468265.83
Average	54480.30	375471.09	2978132.76		45110.57	301811.41	2594435.83
Recovery (%)	82.80	80.38	87.12				
Despropionyl <i>para</i>-fluorofentanyl							
	Set #1				Set #2		
Trial no.	Low:	Med	High		Low	Med	High
1	32789.88	314597.61	2611783.53		44611.95	298602.77	2379969.31
2	32015.33	308942.79	2020791.13		42323.78	262062.45	2461242.98
3	32208.91	279852.22	2524003.09		48160.70	302515.48	2219544.67
Average	32338.04	301130.88	2385525.92		45032.14	287726.90	2353585.65
Recovery (%)	139.25	95.55	98.66				
(±)-<i>cis</i>-3-Methyl fentanyl							
	Set 1				Set 2		
Trial no.	Low	Med	High		Low	Med	High
1	29995.09	191506.88	1699602.14		20491.67	156986.46	1478382.82
2	27567.04	188660.85	1677071.23		21452.07	141056.37	1509950.96
3	30375.57	168344.98	1696079.64		23994.77	155435.24	1401345.31
Average	29312.57	182837.57	1690917.67		21979.50	151159.36	1463226.36
Recovery (%)	74.98	82.67	86.53				
Butyryl/Isobutyryl fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	43402.19	351851.45	2988080.64		39780.73	286850.10	2561000.97
2	45696.44	347785.05	2928941.15		40474.67	250218.59	2732200.06
3	45562.36	310423.46	2915792.27		41938.47	284702.64	2492138.07
Average	44887.00	336686.65	2944271.35		40731.29	273923.77	2595113.03
Recovery (%)	90.74	81.36	88.14				
Carfentanil							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	26402.18	190896.65	1252684.53		20082.97	141783.79	1156826.21
2	26772.94	193793.45	1293548.17		19522.51	125922.13	1232173.90
3	27415.09	164202.38	1352347.09		21596.45	150266.11	1127003.16
Average	26863.40	182964.16	1299526.60		20400.64	139324.01	1172001.09
Recovery (%)	75.94	76.15	90.19				
<i>para</i>-Fluorobutyryl fentanyl/FIBF							

	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	56583.98	381385.01	3052026.12		40598.62	312539.77	2654829.38
2	58804.88	378836.24	2952835.55		39133.46	276976.56	2748411.38
3	55929.45	348080.56	3027438.83		46283.60	320551.42	2534921.27
Average	57106.10	369433.94	3010766.83		42005.23	303355.92	2646054.01
Recovery (%)	73.56	82.11	87.89				
Sufentanil							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	23138.46	154831.15	1361946.43		16366.94	120199.93	1242166.50
2	23670.37	148236.77	1328720.78		15683.01	110473.97	1269157.94
3	24917.06	133487.14	1383698.29		19433.81	121634.57	1138826.22
Average	23908.63	145518.35	1358121.83		17161.25	117436.15	1216716.88
Recovery (%)	71.78	80.70	89.59				
Valeryl fentanyl							
	Set 1				Set 2		
Trial no.	Low	Med	High		Low	Med	High
1	56843.33	462938.62	3830025.05		51047.18	370795.17	3328155.43
2	61663.39	453705.57	3921682.44		50457.26	327512.59	3497367.43
3	59012.31	424183.71	3828719.32		58040.14	369396.01	3238100.13
Average	59173.01	446942.63	3860142.27		53181.53	355901.26	3354541.00
Recovery (%)	89.87	79.63	86.90				

Table S3. Ionization suppression and enhancement percentages for $N = 22$ illicitly manufactured fentanyl (IMFs) analogues at low (0.35 ng mL^{-1}), medium (2.5 ng mL^{-1}), and high control concentrations (25.0 ng mL^{-1}). Set #1 and #2 refer to regular and neat quality standards, respectively.

Norfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	16183.09	112346.21	1040696.84		17631.48	140730.77	1000604.93
2	16400.53	110543.30	1001399.58		18047.00	137202.97	829527.44
3	15576.46	120348.17	1069657.21		18557.65	130331.00	1175213.13
Average	16053.36	114412.56	1037251.21		18078.71	136088.25	1001781.83
Matrix Effect (%)	112.62	118.95	96.58				
Furanyl norfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	4565.76	26311.47	282170.68		4590.15	31704.02	230975.70
2	4934.86	27258.58	196048.07		5487.52	28719.55	212752.81
3	4379.86	27359.57	295672.89		5446.13	27056.84	304847.16
Average	4626.83	26976.54	257963.88		5174.60	29160.14	249525.22
Matrix Effect (%)	111.84	108.09	96.73				
Remifentanil acid							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	1912.22	13115.17	148736.10		2089.06	16561.54	167171.23
2	2044.45	12727.17	158545.18		2334.54	16266.17	157158.90
3	1799.16	13111.16	146491.63		2339.62	15863.99	155740.28
Average	1918.61	12984.50	151257.64		2254.40	16230.56	160023.47
Matrix Effect (%)	117.50	125.00	105.80				
Butyryl norfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	17944.00	133950.12	1168174.85		19330.65	177026.12	1153778.73
2	19068.89	136111.96	1178733.62		21813.09	175292.19	934465.45
3	18347.13	146972.56	1186049.58		20551.76	168768.33	1307357.39
Average	18453.34	139011.55	1177652.68		20565.17	173695.55	1131867.19
Matrix Effect (%)	111.44	124.95	96.11				
Remifentanil							

	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	3698.61	24148.82	245580.98	3398.58	25879.27	247196.84
2	4110.52	31187.58	258044.21	3631.82	26098.03	251472.03
3	3395.64	30361.85	270170.79	3855.28	23094.96	262547.87
Average	3734.92	28566.08	257931.99	3628.56	25024.09	253738.91
Matrix Effect (%)	97.15	87.60	98.37			
Acetyl Fentanyl						
	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	32900.81	272480.22	2696875.16	39163.01	307276.32	2737509.47
2	38733.20	274894.98	2701173.97	39599.84	295551.80	2686140.35
3	38518.67	280970.95	2735233.43	41336.33	279207.70	2690796.28
Average	36717.56	276115.38	2711094.19	40033.06	294011.94	2704815.37
Matrix Effect (%)	109.03	106.48	99.77			
Alfentanil						
	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	16619.67	112384.98	1045263.57	16613.52	111052.10	1069888.51
2	18109.61	118183.23	1046701.52	16148.05	106762.53	1091804.94
3	17756.09	119055.84	1092001.13	16345.64	96063.01	1030880.31
Average	17495.12	116541.35	1061322.07	16369.07	104625.88	1064191.25
Matrix Effect (%)	93.56	89.78	100.27			
AH7921						
	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	6566.28	48615.06	476009.13	5425.16	41519.09	171578.91
2	7633.34	48108.20	473702.01	5448.88	38275.23	187181.59
3	6878.39	47187.37	467335.92	5587.03	35003.78	317460.45
Average	7026.00	47970.21	472349.02	5487.02	38266.03	225406.98
Matrix Effect (%)	78.10	79.77	47.72			
U-47700						
	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	9035.57	60676.63	598626.36	8789.37	69172.60	663875.37
2	8841.10	60748.85	589120.64	7814.93	65002.70	675335.12
3	9052.78	61807.71	555878.02	9579.90	60507.01	611724.71

Average	8976.48	61077.73	581208.34		8728.06	64894.10	650311.73
Matrix Effect (%)	97.23	106.25	111.89				
Acetyl fentanyl 4-methylphenethyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	48878.21	329056.56	2764066.69		43670.09	362857.36	3004272.27
2	49041.99	317459.70	2771548.48		49639.28	358218.25	2896594.04
3	46270.71	329783.06	2784287.37		52373.23	325855.55	2865642.66
Average	48063.64	325433.11	2773300.84		48560.87	348977.05	2922169.66
Matrix Effect (%)	101.03	107.23	105.37				
Acrylfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	43882.58	308992.39	2536934.84		46580.17	341583.14	2646397.46
2	46517.72	293356.95	2596363.31		46952.36	337096.36	2475688.79
3	44189.23	305741.89	2595711.68		48849.41	310714.29	2705321.61
Average	44863.18	302697.08	2576336.61		47460.65	329797.93	2609135.95
Matrix Effect (%)	105.79	108.95	101.27				
Fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	35121.93	243862.09	2277257.74		36202.96	270883.19	2119693.09
2	36338.33	272959.43	2236877.77		35742.90	270642.79	2000594.92
3	37012.04	266771.73	2381041.33		36708.57	245358.38	2271119.18
Average	36157.43	261197.75	2298392.28		36218.14	262294.79	2130469.06
Matrix Effect (%)	100.17	100.42	92.69				
para-Methoxyfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	27910.62	182350.93	1495982.69		27904.20	216676.89	1614255.82
2	28307.99	187967.00	1560701.40		28721.90	201953.30	1627239.48
3	27175.06	188179.96	1535007.01		27416.76	187643.49	1650187.28
Average	27797.89	186165.96	1530563.70		28014.29	202091.22	1630560.86
Matrix Effect (%)	100.78	108.55	106.53				
4ANPP							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High

1	250705.82	1588320.59	10328356.33		166174.74	1261945.01	8907584.39
2	258303.65	1581159.69	10153768.64		164486.85	1336291.01	7977401.72
3	243397.40	1581703.72	10159658.74		153677.98	1223611.13	9025869.40
Average	250802.29	1583728.00	10213927.90		161446.52	1273949.05	8636951.83
Matrix Effect (%)	64.37	80.44	84.56				
Furanyl Fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	49598.48	349942.60	2901533.55		52810.26	387507.42	2941574.99
2	53154.12	346954.45	2836640.95		55872.20	383238.32	2976957.89
3	51906.40	357217.84	2887911.90		54758.43	355667.52	3015865.39
Average	51553.00	351371.63	2875362.13		54480.30	375471.09	2978132.76
Matrix Effect (%)	105.68	106.86	103.57				
Despropionyl <i>para</i>-fluorofentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	55310.89	410215.90	3346009.48		32789.88	314597.61	2611783.53
2	59539.77	417208.83	3306663.71		32015.33	308942.79	2020791.13
3	54462.41	429930.16	3259141.25		32208.91	279852.22	2524003.09
Average	56437.69	419118.30	3303938.15		32338.04	301130.88	2385525.92
Matrix Effect (%)	57.30	71.85	72.20				
(±)-<i>cis</i>-3-Methyl fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	24054.35	166202.47	1583431.31		29995.09	191506.88	1699602.14
2	27032.87	169935.48	1605989.18		27567.04	188660.85	1677071.23
3	28262.03	170303.84	1617468.23		30375.57	168344.98	1696079.64
Average	26449.75	168813.93	1602296.24		29312.57	182837.57	1690917.67
Matrix Effect (%)	110.82	108.31	105.53				
Butyryl/Isobutyryl Fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	45422.09	314101.40	2930769.08		43402.19	351851.45	2988080.64
2	43811.81	330407.44	2863257.61		45696.44	347785.05	2928941.15
3	43617.99	318274.99	2892301.46		45562.36	310423.46	2915792.27
Average	44283.97	320927.94	2895442.71		44887.00	336686.65	2944271.35
Matrix Effect (%)	101.36	104.91	101.69				

Carfentanil							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	29581.26	190158.75	1361256.73		26402.18	190896.65	1252684.53
2	28035.22	185363.84	1327128.00		26772.94	193793.45	1293548.17
3	28947.85	186650.17	1340755.27		27415.09	164202.38	1352347.09
Average	28854.78	187390.92	1343046.67		26863.40	182964.16	1299526.60
Matrix Effect (%)	93.10	97.64	96.76				
para-Fluorobutyryl/FIBF							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	54002.16	342912.34	2955695.73		56583.98	381385.01	3052026.12
2	56611.46	339053.46	2918068.58		58804.88	378836.24	2952835.55
3	58760.62	353647.81	2923275.93		55929.45	348080.56	3027438.83
Average	56458.08	345204.54	2932346.74		57106.10	369433.94	3010766.83
Matrix Effect (%)	101.15	107.02	102.67				
Sufentanil							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	21378.68	141092.61	1353710.93		23138.46	154831.15	1361946.43
2	23467.13	133429.58	1365799.90		23670.37	148236.77	1328720.78
3	21426.18	137804.06	1362923.38		24917.06	133487.14	1383698.29
Average	22090.66	137442.08	1360811.40		23908.63	145518.35	1358121.83
Matrix Effect (%)	108.23	105.88	99.80				
Valeryl Fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	59939.19	373667.31	3834026.52		56843.33	462938.62	3830025.05
2	62192.36	419395.23	3638907.11		61663.39	453705.57	3921682.44
3	60773.11	434846.52	3925964.13		59012.31	424183.71	3828719.32
Average	60968.22	409303.02	3799632.58		59173.01	446942.63	3860142.27
Matrix Effect (%)	97.06	109.20	101.59				

Table S4. Process efficiency percentages for $N = 22$ illicitly manufactured fentanyl (IMFs) analogues at low (0.35 ng mL^{-1}), medium (2.5 ng mL^{-1}), and high control concentrations (25.0 ng mL^{-1}). Set #1 and #2 refer to neat quality standards and regular extractions, respectively.

Norfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	16183.09	112346.21	1040696.84		13814.81	108589.51	894437.11
2	16400.53	110543.30	1001399.58		14056.68	89730.02	960464.26
3	15576.46	120348.17	1069657.21		16278.87	101081.64	872363.61
Average	16053.36	114412.56	1037251.21		14716.78	99800.39	909088.33
Process Efficiency (%)	91.67	87.23	87.64				
Furanyl norfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	4565.76	26311.47	282170.68		2561.18	25373.77	203161.69
2	4934.86	27258.58	196048.07		2868.62	15808.62	254305.58
3	4379.86	27359.57	295672.89		3643.37	17184.88	197428.08
Average	4626.83	26976.54	257963.88		3024.39	19455.76	218298.45
Process Efficiency (%)	65.37	72.12	84.62				
Remifentanil acid							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	1912.22	13115.17	148736.10		888.60	5835.51	50776.07
2	2044.45	12727.17	158545.18		800.58	4657.90	52331.12
3	1799.16	13111.16	146491.63		885.15	5417.15	49659.41
Average	1918.61	12984.50	151257.64		858.11	5303.52	50922.20
Process Efficiency (%)	44.73	40.84	33.67				
Butyryl norfentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	17944.00	133950.12	1168174.85		18740.57	135942.06	1035652.82
2	19068.89	136111.96	1178733.62		17835.67	118133.80	1076581.04
3	18347.13	146972.56	1186049.58		21253.04	129095.32	992372.29
Average	18453.34	139011.55	1177652.68		19276.43	127723.73	1034868.72
Process	104.46	91.88	87.88				

Efficiency (%)						
Remifentanil						
	Set #1				Set #2	
Trial no.	Low	Med	High		Low	Med
1	3698.61	24148.82	245580.98		2769.84	19518.17
2	4110.52	31187.58	258044.21		2732.97	16166.15
3	3395.64	30361.85	270170.79		2863.09	20413.23
Average	3734.92	28566.08	257931.99		2788.64	18699.18
Process Efficiency (%)	74.66	65.46	79.25			
Acetyl Fentanyl						
	Set #1				Set #2	
Trial no.	Low	Med	High		Low	Med
1	32900.81	272480.22	2696875.16		32202.32	242612.47
2	38733.20	274894.98	2701173.97		31485.04	208628.84
3	38518.67	280970.95	2735233.43		35728.32	246516.70
Average	36717.56	276115.38	2711094.19		33138.56	232586.00
Process Efficiency (%)	90.25	84.24	86.20			
Alfentanil						
	Set #1				Set #2	
Trial no.	Low	Med	High		Low	Med
1	16619.67	112384.98	1045263.57		9410.82	83578.37
2	18109.61	118183.23	1046701.52		10494.02	68625.62
3	17756.09	119055.84	1092001.13		11468.14	85460.45
Average	17495.12	116541.35	1061322.07		10457.66	79221.48
Process Efficiency (%)	59.77	67.98	77.80			
AH7921						
	Set #1				Set #2	
Trial no.	Low	Med	High		Low	Med
1	6566.28	48615.06	476009.13		4987.49	30028.21
2	7633.34	48108.20	473702.01		4626.44	29397.38
3	6878.39	47187.37	467335.92		4828.00	33563.29
Average	7026.00	47970.21	472349.02		4813.98	30996.29
Process Efficiency (%)	68.52	64.62	57.70			
U-47700						

	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	9035.57	60676.63	598626.36	7419.27	55748.60	525822.70
2	8841.10	60748.85	589120.64	7911.76	51056.55	554450.77
3	9052.78	61807.71	555878.02	8857.27	53830.68	501931.10
Average	8976.48	61077.73	581208.34	8062.76	53545.28	527401.53
Process Efficiency (%)	89.82	87.67	90.74			
Acetyl fentanyl 4-methylphenethyl						
	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	48878.21	329056.56	2764066.69	39273.94	294742.30	2564883.21
2	49041.99	317459.70	2771548.48	40145.21	270667.02	2671448.69
3	46270.71	329783.06	2784287.37	46585.53	276548.83	2533831.52
Average	48063.64	325433.11	2773300.84	42001.56	280652.71	2590054.47
Process Efficiency (%)	87.39	86.24	93.39			
Acrylfentanyl						
	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	43882.58	308992.39	2536934.84	37202.88	281712.87	2428913.55
2	46517.72	293356.95	2596363.31	37972.97	242770.34	2537661.55
3	44189.23	305741.89	2595711.68	43752.84	287579.53	2351749.58
Average	44863.18	302697.08	2576336.61	39642.89	270687.58	2439441.56
Process Efficiency (%)	88.36	89.43	94.69			
Fentanyl						
	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	35121.93	243862.09	2277257.74	29709.08	227320.04	2041829.06
2	36338.33	272959.43	2236877.77	29167.22	198024.25	2134377.46
3	37012.04	266771.73	2381041.33	35764.60	218804.82	1996871.66
Average	36157.43	261197.75	2298392.28	31546.97	214716.37	2057692.73
Process Efficiency (%)	87.25	82.20	89.53			
para-Methoxyfentanyl						
	Set #1			Set #2		
Trial no.	Low	Med	High	Low	Med	High
1	27910.62	182350.93	1495982.69	22788.33	171129.57	1444155.49

2	28307.99	187967.00	1560701.40		22972.15	154837.11	1453530.64
3	27175.06	188179.96	1535007.01		28583.31	181306.70	1446219.28
Average	27797.89	186165.96	1530563.70		24781.26	169091.13	1447968.47
Process Efficiency (%)	89.15	90.83	94.60				
4ANPP							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	250705.82	1588320.59	10328356.33		190178.80	1229445.02	8369358.55
2	258303.65	1581159.69	10153768.64		180041.98	1076604.71	8607365.57
3	243397.40	1581703.72	10159658.74		202286.59	1208246.04	7867551.53
Average	250802.29	1583728.00	10213927.90		190835.79	1171431.92	8281425.22
Process Efficiency (%)	76.09	73.97	81.08				
Furanyl Fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	49598.48	349942.60	2901533.55		43119.60	311988.90	2601610.85
2	53154.12	346954.45	2836640.95		42896.47	273618.10	2713430.81
3	51906.40	357217.84	2887911.90		49315.64	319827.23	2468265.83
Average	51553.00	351371.63	2875362.13		45110.57	301811.41	2594435.83
Process Efficiency (%)	87.50	85.90	90.23				
Despropionyl <i>para</i>-fluorofentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	55310.89	410215.90	3346009.48		44611.95	298602.77	2379969.31
2	59539.77	417208.83	3306663.71		42323.78	262062.45	2461242.98
3	54462.41	429930.16	3259141.25		48160.70	302515.48	2219544.67
Average	56437.69	419118.30	3303938.15		45032.14	287726.90	2353585.65
Process Efficiency (%)	79.79	68.65	71.24				
(±)-<i>cis</i>-3-Methyl fentanyl							
	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	24054.35	166202.47	1583431.31		20491.67	156986.46	1478382.82
2	27032.87	169935.48	1605989.18		21452.07	141056.37	1509950.96
3	28262.03	170303.84	1617468.23		23994.77	155435.24	1401345.31
Average	26449.75	168813.93	1602296.24		21979.50	151159.36	1463226.36

Process Efficiency (%)	83.10	89.54	91.32			
Butyryl/Isobutyryl Fentanyl						
	Set #1				Set #2	
Trial no.	Low	Med	High		Low	Med
1	45422.09	314101.40	2930769.08		39780.73	286850.10
2	43811.81	330407.44	2863257.61		40474.67	250218.59
3	43617.99	318274.99	2892301.46		41938.47	284702.64
Average	44283.97	320927.94	2895442.71		40731.29	273923.77
Process Efficiency (%)	91.98	85.35	89.63			
Carfentanil						
	Set #1				Set #2	
Trial no.	Low	Med	High		Low	Med
1	29581.26	190158.75	1361256.73		20082.97	141783.79
2	28035.22	185363.84	1327128.00		19522.51	125922.13
3	28947.85	186650.17	1340755.27		21596.45	150266.11
Average	28854.78	187390.92	1343046.67		20400.64	139324.01
Process Efficiency (%)	70.70	74.35	87.26			
para-Fluorobutyryl/FIBF						
	Set #1				Set #2	
Trial no.	Low	Med	High		Low	Med
1	54002.16	342912.34	2955695.73		40598.62	312539.77
2	56611.46	339053.46	2918068.58		39133.46	276976.56
3	58760.62	353647.81	2923275.93		46283.60	320551.42
Average	56458.08	345204.54	2932346.74		42005.23	303355.92
Process Efficiency (%)	74.40	87.88	90.24			
Sufentanil						
	Set #1				Set #2	
Trial no.	Low	Med	High		Low	Med
1	21378.68	141092.61	1353710.93		16366.94	120199.93
2	23467.13	133429.58	1365799.90		15683.01	110473.97
3	21426.18	137804.06	1362923.38		19433.81	121634.57
Average	22090.66	137442.08	1360811.40		17161.25	117436.15
Process Efficiency (%)	77.69	85.44	89.41			
Valeryl Fentanyl						

	Set #1				Set #2		
Trial no.	Low	Med	High		Low	Med	High
1	59939.19	373667.31	3834026.52		51047.18	370795.17	3328155.43
2	62192.36	419395.23	3638907.11		50457.26	327512.59	3497367.43
3	60773.11	434846.52	3925964.13		58040.14	369396.01	3238100.13
Average	60968.22	409303.02	3799632.58		53181.53	355901.26	3354541.00
Process Efficiency (%)	87.23	86.95	88.29				

Acquisition Method Info

Method Name fentanalogmeth.m
Method Path D:\MassHunter\Methods\fentanalogmeth.m
Method Description Fentanyl Analogs MRM

Device List

HiP Sampler
 Binary Pump
 Column Comp.
 QQQ

MS QQQ Mass Spectrometer

Ion Source ESI **Tune File** atunes.TUNE.XML
Stop Mode No Limit/As Pump **Stop Time (min)** 2.0
Time Filter On **Time Filter Width (min)** 0.07
LC->Waste Pre Row N/A **LC->Waste Post Row** N/A

Time Segments

Index	Start Time (min)	Scan Type	Ion Mode	Div Valve	Delta EMV	Store	Cycle Time (ms)	Triggered ?	MRM Repeats
1	0.5	DynamicMRM	ESI	To MS	300	Yes	500	No	3

Time Segment 1

Scan Segments

Cpd Name	ISTD?	Prec Ion	MS1 Res	Prod Ion	MS2 Res	Frag (V)	CE (V)	Cell Acc (V)	Ret Time (min)	Ret Window	Polarity
1-3-Methylfentanyl	No	351.51	Unit/Enh (6490)	202.1	Unit/Enh (6490)	150	20	7	9.511	1	Positive
1-3-Methylfentanyl	No	351.51	Unit/Enh (6490)	105	Unit/Enh (6490)	150	48	7	9.511	1	Positive
4ANPP	No	281.4	Unit/Enh (6490)	105.1	Unit/Enh (6490)	116	36	7	9.39	1	Positive
4ANPP	No	281.4	Unit/Enh (6490)	77.2	Unit/Enh (6490)	116	76	7	9.39	1	Positive
4ANPP	No	281.4	Unit/Enh (6490)	51.3	Unit/Enh (6490)	116	124	7	9.39	1	Positive
Acetyl Fent 4-meth	No	337.5	Unit/Enh (6490)	119	Unit/Enh (6490)	136	36	7	9.224	1	Positive
Acetyl Fent 4-meth	No	337.5	Unit/Enh (6490)	91.1	Unit/Enh (6490)	136	72	7	9.224	1	Positive
Acetyl Fentanyl	No	323	Unit/Enh (6490)	188	Unit/Enh (6490)	141	20	7	8.8	1	Positive
Acetyl Fentanyl	No	323	Unit/Enh (6490)	105	Unit/Enh (6490)	141	40	7	8.8	1	Positive
AcetylFentanyl-13C6	Yes	329.4	Unit/Enh (6490)	105	Unit/Enh (6490)	136	44	7	8.864	1	Positive
AcetylFentanyl-13C6	Yes	329.4	Unit/Enh (6490)	77.1	Unit/Enh (6490)	136	96	7	8.864	1	Positive
Acrylfentanyl	No	335.5	Unit/Enh (6490)	105	Unit/Enh (6490)	141	44	7	9.229	1	Positive
Acrylfentanyl	No	335.5	Unit/Enh (6490)	77.1	Unit/Enh (6490)	141	92	7	9.229	1	Positive
Acrylfentanyl	No	335.5	Unit/Enh (6490)	51.2	Unit/Enh (6490)	141	140	7	9.229	1	Positive
AH7921	No	329	Unit/Enh (6490)	284	Unit/Enh (6490)	111	20	7	8.864	1	Positive
AH7921	No	329	Unit/Enh (6490)	95.1	Unit/Enh (6490)	111	36	7	8.864	1	Positive
Alfentanil	No	417.5	Unit/Enh (6490)	165	Unit/Enh (6490)	131	36	7	8.852	1	Positive
Alfentanil	No	417.5	Unit/Enh (6490)	99	Unit/Enh (6490)	131	40	7	8.852	1	Positive
Alfentanil	No	417.5	Unit/Enh (6490)	77.1	Unit/Enh (6490)	131	100	7	8.852	1	Positive
Butyryl Fent/Isobut	No	351.2	Unit/Enh (6490)	188.1	Unit/Enh (6490)	146	24	7	9.5	1	Positive

Acquisition Method Report



yl Butyl Fent/Isobut yl	No	351.2	Unit/Enh (6490)	105.1	Unit/Enh (6490)	146	48	7	9.5	1	Positive
yl Butyl norfentanyl	No	247.3	Unit/Enh (6490)	94	Unit/Enh (6490)	106	32	7	8.135	1	Positive
yl Butyl norfentanyl	No	247.3	Unit/Enh (6490)	84.1	Unit/Enh (6490)	106	16	7	8.135	1	Positive

Acquisition Method Report



Cpd Name	ISTD?	Prec Ion	MS1 Res	Prod Ion	MS2 Res	Frag (V)	CE (V)	Cell Acc (V)	Ret Time (min)	Ret Window	Polarity
Butyryl norfentanyl	No	247.3	Unit/Enh (6490)	55.2	Unit/Enh (6490)	106	44	7	8.135	1	Positive
Carfentanil	No	395.21	Unit/Enh (6490)	113	Unit/Enh (6490)	131	36	7	9.455	1	Positive
Carfentanil	No	395.21	Unit/Enh (6490)	105	Unit/Enh (6490)	131	56	7	9.455	1	Positive
Carfentanil	No	395.21	Unit/Enh (6490)	77.1	Unit/Enh (6490)	131	112	7	9.455	1	Positive
Desprop Fluorofent	No	299.4	Unit/Enh (6490)	105	Unit/Enh (6490)	111	36	7	9.463	1	Positive
Desprop Fluorofent	No	299.4	Unit/Enh (6490)	77.1	Unit/Enh (6490)	111	88	7	9.463	1	Positive
Desprop Fluorofent	No	299.4	Unit/Enh (6490)	51.2	Unit/Enh (6490)	111	88	7	9.463	1	Positive
Fentanyl	No	337.5	Unit/Enh (6490)	188	Unit/Enh (6490)	131	20	7	9.224	1	Positive
Fentanyl	No	337.5	Unit/Enh (6490)	105	Unit/Enh (6490)	131	44	7	9.224	1	Positive
Fentanyl-D5	Yes	342.5	Unit/Enh (6490)	105	Unit/Enh (6490)	141	44	7	9.22	1	Positive
Fentanyl-D5	Yes	342.5	Unit/Enh (6490)	77.1	Unit/Enh (6490)	141	100	7	9.22	1	Positive
Furanyl Fentanyl	No	375.1	Unit/Enh (6490)	188.2	Unit/Enh (6490)	125	25	7	9.423	1	Positive
Furanyl Fentanyl	No	375.1	Unit/Enh (6490)	105	Unit/Enh (6490)	125	40	7	9.423	1	Positive
Furanyl norfentanyl	No	271.4	Unit/Enh (6490)	95	Unit/Enh (6490)	106	44	7	7.983	1	Positive
Furanyl norfentanyl	No	271.4	Unit/Enh (6490)	84.1	Unit/Enh (6490)	106	16	7	7.983	1	Positive
Norfentanyl I	No	233.4	Unit/Enh (6490)	94	Unit/Enh (6490)	106	36	7	7.77	1	Positive
Norfentanyl I	No	233.4	Unit/Enh (6490)	84.1	Unit/Enh (6490)	106	16	7	7.77	1	Positive
Norfentanyl I	No	233.4	Unit/Enh (6490)	55.2	Unit/Enh (6490)	106	44	7	7.77	1	Positive
Norfentanyl I-D5	Yes	238.4	Unit/Enh (6490)	84.1	Unit/Enh (6490)	106	16	7	7.651	1	Positive
Norfentanyl I-D5	Yes	238.4	Unit/Enh (6490)	55.2	Unit/Enh (6490)	106	44	7	7.651	1	Positive
para-Fluorobutyryl/FIBF	No	369.24	Unit/Enh (6490)	188.1	Unit/Enh (6490)	141	24	7	9.523	1	Positive
para-Fluorobutyryl/FIBF	No	369.24	Unit/Enh (6490)	105.1	Unit/Enh (6490)	141	44	7	9.523	1	Positive
para-Fluorobutyryl/FIBF	No	369.24	Unit/Enh (6490)	77.1	Unit/Enh (6490)	141	108	7	9.523	1	Positive
para-Methoxyfentanyl	No	367.6	Unit/Enh (6490)	105	Unit/Enh (6490)	136	44	7	9.357	1	Positive
para-Methoxyfentanyl	No	367.6	Unit/Enh (6490)	77.1	Unit/Enh (6490)	136	108	7	9.357	1	Positive
para-Methoxyfentanyl	No	367.6	Unit/Enh (6490)	51.2	Unit/Enh (6490)	136	160	7	9.357	1	Positive
Remifentanyl	No	377.46	Unit/Enh (6490)	345	Unit/Enh (6490)	25	15	7	8.456	1	Positive
Remifentanyl	No	377.46	Unit/Enh (6490)	317	Unit/Enh (6490)	25	15	7	8.456	1	Positive
Remifentanyl metab.	No	363.4	Unit/Enh (6490)	81.1	Unit/Enh (6490)	111	44	7	8.048	1	Positive
Remifentanyl metab.	No	363.4	Unit/Enh (6490)	53.2	Unit/Enh (6490)	111	72	7	8.048	1	Positive
Sufentanil	No	387.56	Unit/Enh (6490)	238.18	Unit/Enh (6490)	121	36	7	9.713	1	Positive
Sufentanil	No	387.56	Unit/Enh (6490)	132	Unit/Enh (6490)	121	36	7	9.713	1	Positive
Sufentanil	No	387.56	Unit/Enh (6490)	111	Unit/Enh (6490)	121	44	7	9.713	1	Positive
U-47700	No	329	Unit/Enh (6490)	204	Unit/Enh (6490)	120	25	7	9.011	1	Positive
U-47700	No	329	Unit/Enh (6490)	81	Unit/Enh (6490)	120	36	7	9.011	1	Positive
Valeryl Fentanyl	No	365.5	Unit/Enh (6490)	105	Unit/Enh (6490)	136	44	7	9.5	1	Positive
Valeryl Fentanyl	No	365.5	Unit/Enh (6490)	77.1	Unit/Enh (6490)	136	112	7	9.5	1	Positive
Valeryl Fentanyl	No	365.5	Unit/Enh (6490)	51.2	Unit/Enh (6490)	136	164	7	9.5	1	Positive

Scan Parameters

Data Stg	Threshold
Centroid	0

Source Parameters

Parameter	Value (+)	Value (-)
Gas Temp (°C)	350	350
Gas Flow (l/min)	12	12
Nebulizer (psi)	60	60
Capillary (V)	4000	4000

Chromatograms

Chrom Type	Label	Offset	Y-Range
TIC	TIC	0	10000000

Instrument Curves

Actual

HiP Sampler

Model: G1367B

Name:
Auxiliary

Draw Speed	200.0 µL/min
Eject Speed	200.0 µL/min
Draw Position Offset	1.0 mm
Wait Time After Drawing	2.0 s
Sample Flush Out Factor	20.0
Vial/Well bottom sensing	No

Injection

Injection Mode	Injection with needle wash
Injection Volume	10.00 µL
Needle Wash	
Needle Wash Location	Flush Port
Wash Time	35.0 s

High throughput

Automatic Delay Volume Reduction	No
Overlapped Injection	
Enable Overlapped Injection	No

Valve Switching

Valve Movements	1
Valve Switch Time 1	
Switch Time 1 Enabled	No
Valve Switch Time 2	
Switch Time 2 Enabled	No
Valve Switch Time 3	
Switch Time 3 Enabled	No
Valve Switch Time 4	
Switch Time 4 Enabled	No

Stop Time

Stoptime Mode	As pump/No limit
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Post Time

Posttime Mode	Off
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Acquisition Method Report



Name:

Binary Pump

Model:

G1312A

Flow 0.400 mL/min
 Low Pressure Limit 0.00 bar
 High Pressure Limit 400.00 bar
 Maximum Flow Gradient 100.000 mL/min²

Stroke A

Automatic Stroke Calculation A Yes

Stroke B

Automatic Stroke Calculation B Yes

Compress A

Compressibility Mode A Compressibility Value Set
 Compressibility A 50 10e-6/bar

Compress B

Compressibility Mode B Compressibility Value Set
 Compressibility B 115 10e-6/bar

Stop Time

Stoptime Mode Time set
 Stoptime 13.50 min

Post Time

Posttime Mode Off

Solvent Composition

	Channel	Name 1	Name 2	Selected	Used	Percent
1	A	amm. formate, formic, water	amm. formate, formic, water	Ch. 2	Yes	90.0 %
2	B	formic, ACN	formic, ACN	Ch. 2	Yes	10.0 %

Timetable

	Time	A	B	Flow	Pressure
1	2.00 min	90.0 %	10.0 %	0.400 mL/min	400.00 bar
2	8.00 min	10.0 %	90.0 %	0.400 mL/min	400.00 bar
3	8.50 min	10.0 %	90.0 %	0.400 mL/min	400.00 bar
4	8.60 min	90.0 %	10.0 %	0.400 mL/min	400.00 bar
5	12.00 min	90.0 %	10.0 %	0.400 mL/min	400.00 bar

Column Comp.

Model:

G1316A

Name:

Valve Position Port 1 -> 6

Left Temperature Control

Temperature Control Mode Temperature Set
 Temperature 40.00 °C

Enable Analysis Left Temperature

Enable Analysis Left Temperature On Yes
 Enable Analysis Left Temperature Value 0.8 °C

Right Temperature Control

Right temperature Control Mode Not Controlled

Enable Analysis Right Temperature

Enable Analysis Right Temperature On Yes
 Enable Analysis Right Temperature Value 0.8 °C

Stop Time

Stoptime Mode As pump/injector

Post Time

Posttime Mode Off